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QUALITY ASSESSMENT OF SOME GARI SAMPLES FROM IWO, OSUN STATE, NIGERIA

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ABSTRACT

Gari, prepared from fermented cassava (*Manihot esculanta Crantz)* can be considered as ready-to eat popular food among all Nigerians regardless of age, gender, education and income. Exposure of gari to the environment during processing and at points of sale, makes the product susceptible to mishandling and contaminations. Microorganisms and possible presence of toxins or metabolites in gari makes it a source of pathogens and consumption could result in foodborne poisoning which is of public health concern. The focus of this study was to evaluate the quality of white and yellow gari sold in Iwo and the resulting eba. Physico-chemical and microbial analyses were carried out using standard methods. Swelling index ranged from (3.10 ± 0.1 to 3.43 ± 0.1); Water absorption capacity ranged from (2.95 ± 0.1 to 3.66 ± 0.2); bulk and tap density ranged from (0.55 ± 0.0 to 0.59 ± 0.0) and (0.60 ± 0.0 to 0.64 ± 0.0) respectively. The pH ranged from (2.86 ± 0.0 to 3.55 ± 0.0). Total Titratable Acidity values ranged from (0.01 ± to 0.02 ± 0.0). Microbial load ranged from (4.1 x 10^6 to 3.48 x 10^7 CFU/g); (3.30 x 10^6 to 1.09 x 10^7 CFU/g); (2.32 x 10^7 to Too Numerous To Count (TNTC)); (9.50 x 10^5 to TNTC) for total viable (TVC), Staphylococcal, Enterobacteriaceae and fungal counts respectively. All gari samples exceeded SON allowable limit for microorganisms, but eba samples had lower microbial (1.6x10^6 to 3.4x10^6 CFU/g); (No growth)) and (4.1x10^6 to 9.5x10^6 CFU/g) for TVC, Enterobacteriaceae and fungal load. Isolated and identified organisms included *Klebsiella* spp., *Enterobacter* spp., *Escherichia coli*, *Staphylococcus aureus and Aspergillus niger*. The study concludes that gari from Iwo markets exceed the allowable microbial limits, contain Food borne pathogens and is of public health concern.

Keywords: Gari; quality; microbial load; food safety.

INTRODUCTION

Cassava (*Manihot esculenta* Crantz) is a perennial shrub with edible starchy roots cultivated in the tropical and sub-tropical regions of the world. A variety of foodstuffs are made from cassava including lafun, fufu and gari. Gari is a granular flour (creamy white or yellow) and a very popular food in West Africa and among all Nigerians. It is commonly consumed as ready to eat food (Oluwole *et al*., 2004). During sales and bargaining, dry gari is tested for general acceptability by consumers. After purchase, it is consumed by soaking in cold water with sugar, coconut, roasted groundnuts, dry fish, or boiled cowpea as complements. It is also consumed as a paste made with hot water and eaten with

vegetable sauce (ARS, 2012). The popularity and consumption of gari without cooking transcends age, gender, education and affluence.

Gari has been the focus of many studies and a lot has been discovered, such as its method of production, as a cottage industry (Aworh, 2008), majorly in villages (Makanjuola *et al.,* 2012), nutritional content (Ojo and Akande, 2013), antinutritional content (Owuamanam et al., 2010) and fermenting microorganisms (Bokanga, 1995; Ijabadeniyi, 2007). There are various sources of microbial contamination in gari (Aguoru *et al.*, 2014). These include the equipment and utensils used in production such as stainless knives, stones, woven baskets, jute bags, clay stoves which are oftentimes not adequately cleaned and can contaminate the product. Other unhygienic practices and dirty environment can increase microbial contamination of gari. In addition, drying on mats; perching of infected vectors such as housefly, insects and mites during storage; transportation; open display in bowls at points of sale (Ogiehor and Ikenebomeh, 2006) and handling with bare hands during trading by both the vendor and the customers, further increase the microbial load (Ogugbue and Obi, 2011). Microbial contamination can be from spoilage or pathogenic microorganisms or both. Spoilage organisms such as *Rhizopus nigricans* hamper the storage life of gari leaving biochemical and sensory changes in it ranging from discoloration to tainting and off flavour development. While the pathogenic microorganisms such as *Staphylococcus aureus* can cause foodborne illnesses in a population of consumers (Jhalka *et al*., 2014) and is of food safety and public health concern Gari is a hygroscopic food product, storage or packaging in an environment of 70% relative humidity is important in order to limit microbial deterioration (Linssen and Roozen, 1994). Gari is prone to mishandling and infestation of vectors during sales and storage. Individuals attempting to assess the quality of garri may also be sources of human pathogens. Since gari is popularly consumed without further preparation by young and old alike, it is an important food vehicle for foodborne diseases and should be of public health concern. Hence, the focus of this study was to assess the quality of some gari samples from Iwo area, and to identify the food borne microorganisms of public health concern there in.

MATERIALS AND METHODS

Materials

A total of five gari samples were purchased and analyzed. Three (2 white and 1 yellow gari), displayed from open markets; 1 unpackaged from producer and 1 packaged from a store in Iwo. Samples were transported within 1 hour of procurement to the Department of Food Science and Technology laboratory, Bowen University. They were labeled and used within 8 hrs for microbial analysis in order to avoid contamination. But for other analyses, samples were stored at ambient (32±2 ^{0}C) temperature and used within a few days.

Methods

pH

About 10 g of sample was homogenized in 100 mL of De-ionized water according to Ogiehor and Ikenebomeh (2005). The free water was decanted and the pH (Mettler Toledo AG 8603, Switzerland) of suspension was read.

Titratable Acidity (TTA)

TTA of all samples was determined by following the method described by AOAC (2005) with slight modification. Approximately 10 g of sample was soaked in 100 mL of De-ionized water for 10 min. About 10 mL of the supernatant was titrated against 0.1M sodium hydroxide using phenolphthalein as an indicator until the appearance of a pink colouration. Total titratable acidity was expressed as percentage of lactic acid.

Swelling index

Swelling index was determined according to Sanni *et al* (2001) method. Briefly, a 100 mL glass 20 mL mark on the cylinder and De-ionized water was added at room (32± 2 °C) temperature to make a suspension up to 100 mL. The cylinder and sample was inverted, homogenized for 2 min and left to stand for additional 3 min. Final volume occupied by the gari was recorded and swelling index was calculated as:

$$\text{Swelling Index} = \frac{V_2}{V_1} \times 100$$

Where V1=the initial volume of gari in the cylinder;
V2 =the final volume of gari in water

Water Absorption Capacity (WAC)

WAC was determined according to Nuwamanya *et al* (2011), with slight modifications. About 1 g of each sample was dissolved in 10 mL of water in a centrifuge tube. The suspension was agitated for 3 min in a Stuart wrist shaker (Bibby Scientific Ltd., Staffordshire, UK), and allowed to stand for additional 10 min and after which it was centrifuged (Uniscope SM 902B, England, UK) for 10 min at 1000 rpm. The supernatant was decanted from the residue, drained for 10 min and weighed. The difference in weight was recorded as water absorbed. The WAC is calculated as:

$$\text{WAC} = \frac{\text{Weight of Water bound}}{\text{Weight of sample}} \times 100$$

Bulk and Packed density

Bulk density was as described by Picker-Freyer and Brink (2006), with slight modification. Approximately 100 g of sample was carefully filled into a pre-weighed 250 mL glass measuring cylinder. The mean value was recorded from triplicate determinations and bulk density was calculated as:

$$\text{Bulk density} = \frac{\text{Weight of sample}}{\text{Loose volume of sample}} \times 100$$

Then, the cylinder was tapped for at least 50 times for a constant volume. The packed volume was recorded and calculated as packed density:

$$\text{Packed density} = \frac{\text{Weight of sample}}{\text{Packed volume of sample}} \times 100$$

Both bulk and tap density were expressed as g/mL.

Preparation of dough (Eba) from gari samples

Dough (eba) samples were prepared by add 150 mL of hot boiling (100 ^{0}C) water to 5 g of gari in a sterile bowl. The unabsorbed water was decanted off and dough was stirred with a sterile spoon, covered and left to cool before microbial analysis.

Microbial Analyses

Microbial load of gari and resulting dough (eba) samples

About 1 g of each gari sample was weighed into 9 mL of peptone water, homogenized and further serially diluted up to 10^{-5} according to Ogbuile *et al* (1998). Using the pour plate method, 1mL of the last dilution was plated aseptically in duplicates on Plate count, MacConkey, Mannitol salt and Potato Dextrose agars (Lab M). Following cooling, the eba samples were analyzed as previously described. The plates were incubated at 37 ^{0}C for 18-24 h and fungal plates at 25 ^{0}C for 72 h. Colonies were enumerated after incubation.

Identification of Isolates

Following enumeration, distinctive morphological characteristics of each colony, as described by (Brown, 2009) were observed. Isolates were purified by repeated sub-culturing for further biochemical studies and identification (Pollack *et al.,* 2002) while fungal isolates were identified based on conider heads, phialides, conidiophores and the presence or absence of foot cells or rhizoids (Samson and Reenca-Hoekstra, 1988; Bounds *et al.,* 1993).

Statistical Analysis

Data collected were analyzed using Statistical Package for Social Sciences (SPSS) IBM version 20, 2011. Analysis of Variance (ANOVA) was used to evaluate significant differences ($p<0.05$). Duncan test was used for the separation of means.

RESULTS AND DISCUSSION

The results for pH, TTA, swelling index and water absorption capacity are reported in Table 1.

pH

The pH of the products ranged from (2.7 ± 0.0 to 3.6 ± 0.0). Gari packaged in LDPE had the least pH of 2.7 while yellow gari had the highest pH of 3.6. The pH of gari samples from this study slightly vary and are significantly ($P<0.05$) different. Furthermore, they are lower than (4.3 to 4.5) reported by Makanju *et al* (2012).

Titratable Acidity (TTA)

The TTA of the products ranged between (0.01 ± 0.0 % and 0.02 ± 0.0 %). There was no significant difference ($P<0.05$) between the samples. TTA values of the gari samples did not meet the minimum TTA specification of (0.6 %) for gari (ARS, 2012) but values are comparable to the reports of (0.03 to 0.04) and (0.01 to 0.04%) (Makanjuola *et al.,* 2012; Ototu *et al.,* 2013).

Swelling Index

The swelling capacity of gari ranged from (3.1 ± 0.1 to 3.4 ± 0.1). Yellow gari had the highest swelling capacity of (3.4 ± 0.1) while white gari from local Retailer 2 had the least swelling capacity of (3.1 ± 0.1).

Water absorption capacity (WAC)

The WAC of the samples ranged from (3.0 ± 0.1 to 3.7 ± 0.2). Overall, there were significant differences ($p<0.05$) between the samples, with LDPE packaged white gari having the highest WAC while Producer (unpackaged gari) had the lowest water absorption capacity.

TABLE 1: Mean values of physico-chemical and functional properties of gari

Sample	pH	TTA (%)	SI (mL)	WAC (%)
R1WG	3.2 ± 0.5^{ab}	0.01 ± 0.0^{a}	$3.1\pm.2^{b}$	$3.1\pm.3^{b}$
R2WG	2.9 ± 0.0^{bc}	0.01 ± 0.0^{a}	$3.1\pm.1^{b}$	$3.4\pm.1^{ab}$
PUWG	3.3 ± 0.0^{ab}	0.01 ± 0.0^{a}	$3.2\pm.1^{b}$	$3.0\pm.1^{c}$
R3YG	3.6 ± 0.0^{a}	0.01 ± 0.0^{a}	$3.4\pm.1^{a}$	$3.2\pm.2^{b}$
LPWG	2.7 ± 0.0^{c}	0.02 ± 0.0^{a}	$3.4\pm.2^{a}$	$3.7\pm.2^{a}$

R1WG= Retailer 1 white gari; R2WG= Retailer 2 white gari; PUWG=Producer unpackaged white gari; R3YG=Retailer 3 yellow gari; and LPWG=LDPE packaged white gari. Values are mean ± SD of 3 replicates. Test values along the same column carrying different superscripts for each parameter are significantly different ($p < 0.05$) according to Duncan test.

Bulk and Packed density

Results for bulk and packed density are presented in Fig 1. Bulk and packed density ranged from (0.55 ± 0.0 to 0.59 ± 0.0) and (0.60 ± 0.0 to 0.64 ± 0.0) respectively. There were significant ($P<0.05$) difference between the samples. R2WG and PUWG had the least bulk density of (0.55 g/mL) while Retailers 1 and 3 (white and yellow gari) had the highest (0.59 g/mL) bulk density. The swelling capacity corresponds with the reports of (3.02 to 3.85) from Otutu *et al* (2013) and (3.16 to 3.51) from Makanjuola *et al* (2012). Good quality gari within the granules. The range of WAC obtained is within the report of Otutu *et al* (2013). The lower the bulk density of any product, the

lower its floatation ability which can lead to product rejection by the consumers due to the product's inability to soak well in water (Sanni *et al*., 2008) or even swell as required. Tap density values ranged from (0.60 to 0.64 g/mL). White gari from Retailer 2 and Producer (unpackaged) white gari differ significantly from the other samples. should have ability to gain three times its size in water (Almazan *et al*., 1987). According to Sanni *et al* (2001; 2005) the swelling index of granules reflect the extent of associative force

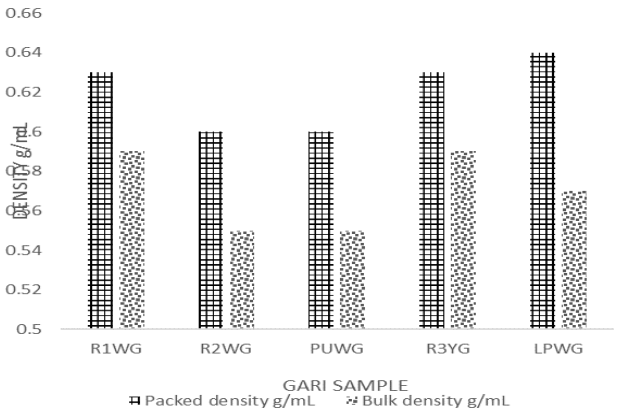

Figure 1: Packed and Bulk density of gari samples.

R1WG= Retailer 1 white gari; R2WG= Retailer 2 white gari; PUWG=Producer unpackaged white gari; R3YG=Retailer 3 yellow gari; and LPWG=LDPE packaged white gari.

Microbial Analyses

In order to assess the microbial quality of gari from various locations and the resulting eba dough samples were subjected to microbial analysis and the results are presented in Table 2. Overall there were growths in all the agars, indicating that all the gari samples were contaminated. Microbial load ranged from ($4.1x10^6$ to $3.5x10^7$ CFU/g); ($3.3x10^6$ to $1.1x10^7$ CFU/g); ($2.3 x10^7$ to TNTC); ($9.5 x 10^5$ to TNTC) for total viable (TVC), Staphylococcal, Enterobacteriaceae and fungal counts respectively. Microbial load for Eba dough ranged from ($1.6x10^6$ to $3.4x10^6$ CFU/g); ($4.1x10^6$ to $9.5x10^6$ CFU/g) for TVC and fungal counts respectively, but no growth was observed for Enterobacteriaceae.

Table 2: Microbial load of soaked un-decanted gari and eba dough sample

Sample	Microbial load (CFU/g) TVC	Staphylococcal Count	Enterobacteriaceae Count	Fungal Count
Soaked not decanted				
R1WG				
R2WG	4.1×10^6	$4.5^c \times 10^6$	$2.7^b \times 10^7$	$9.5^d \times 10^5$
PUWG	$7.4^d \times 10^6$	$1.0^a \times 10^7$	$2.4^c \times 10^7$	$2.0^c \times 10^6$
R3YG	$1.2^c \times 10^7$	$6.9^b \times 10^6$	$2.3^d \times 10^7$	$3.0^b \times 10^6$
LPWG	$3.4^b \times 10^7$	$1.1^a \times 10^7$	TNTC*	TNTC
Eba dough	$3.5^a \times 10^7$	$3.3^c \times 10^6$	TNTC	TNTC
R1WG	$2.7^b \times 10^6$	ND	NG	$4.1^d \times 10^6$
R2WG	$3.4^a \times 10^6$	ND	**NG**	$6.7^c \times 10^6$
PUWG	$2.0^c \times 10^6$	ND	NG	$8.4^b \times 10^6$
R3YG	$1.6^d \times 10^6$	ND	**NG**	$9.5^a \times 10^6$
LPWG	$1.9^c \times 10^6$	ND	**NG**	$6.2^c \times 10^6$

R1WG= Retailer 1 white gari; R2WG= Retailer 2 white gari; PUWG=Producer unpackaged white gari;

R3YG=Retailer 3 yellow gari; and LPWG=LDPE packaged white gari. TNTC* = Too numerous to count; ND[1]=Not determined; NG[2] = No growth. Duncan separation, means with same alphabets are not different ($p<.05$) in each row.

The microbiological quality exceeds the limits of TVC (10^3 CFU/g); Staphylococcal (10^2 CFU/g); *Escherichia coli*, *Salmonella*, *Coliforms and Bacillus cereus* (0 CFU/g) and fungi (10^3 CFU/g) count (ARS, 2012). LDPE packaged white gari had the highest ($3.5x\ 10^7$ CFU/g) TVC; Retailer 3 yellow gari had the highest (1.1×10^7 CFU/g) Staphylococcal; LDPE packaged and yellow gari had too numerous to count (TNTC) for Enterobacteriaceae and fungal count. Microbial load for soaked and decanted gari samples were also determined and found to have significantly lower microbial content compared to soaked samples that were not decanted (data not included). The high incidence of both bacteria and fungi contamination in the samples could be due to poor manufacturing and poor hygienic practices of preparers, environment and storage as well as the poor post-harvest handling of the products. High microbial load of gari in LDPE package in this study suggest that LDPE may not be an effective packaging material for gari and could be attributable to relative permeability of microorganisms, oxygen, carbon dioxide and water vapour through the LDPE packaging material. Permeability characteristics and oxygen transfer rate are the cause for progressive microbial count (Amadi and Adebola, 2008; Efiuvwevwere and Uwanogho, 1990; Paine, 1992). Furthermore, the report of Ogiehor and Ikenebomeh (2006) also reflect an increase in the microbial load of gari packaged in LDPE as storage time increases.

Identification of Isolates

Observation of colony morphology, Gram staining and biochemical analyses of the isolates from gari samples show that the tentative identification of the bacteria isolates were *Klebsiella* spp., *Enterobacter*, *Escherichia coli*, *Bacillus* spp., *Staphylococcus* aureus. Fungi isolates from gari from this study included *Aspergillus niger*, *Penicillium* spp., *Geotrichum* spp., (Table 3). The frequency of isolation of the types of microorganisms from the gari samples used in this study is presented in Fig 2. The organisms isolated and identified in this study are similar to previous works of gari (Ogugbue *et al*., 2011; Ogugbue and Obi, 2011). The presence of spore formers namely: *Bacillus subtilis*, *Penicillium* spp., *Aspergillus fumigatus*, *Rhizopus nigricans*, *Mucor* spp. *and Aspergillus niger*, *Bacillus* spp. and *Geotrichum candidum* isolated from the samples may be attributed to the survival and resuscitation of spores after gari making process (Ogugbue *et al*., 2011). The presence of pathogens such as *E. coli*, *Klebsiella* and staphylococcal strains in gari samples indicate poor post processing handling of samples in the open market, packaged as well as that which was obtained directly from the Producer. These aforementioned pathogens have been implicated as causative agents of foodborne disease outbreaks (Ajayi *et al.*, 2011).

Other organisms can cause overall deterioration, loss in food quality and safety of the product, thus, leading to economic losses, consumers' dissatisfaction and food poisoning in a population, particularly if gari is consistently consumed without decanting the initial soak water. Soaking and consuming gari in unclean or untreated water will undoubtedly increase the diversity and load of microbial content.

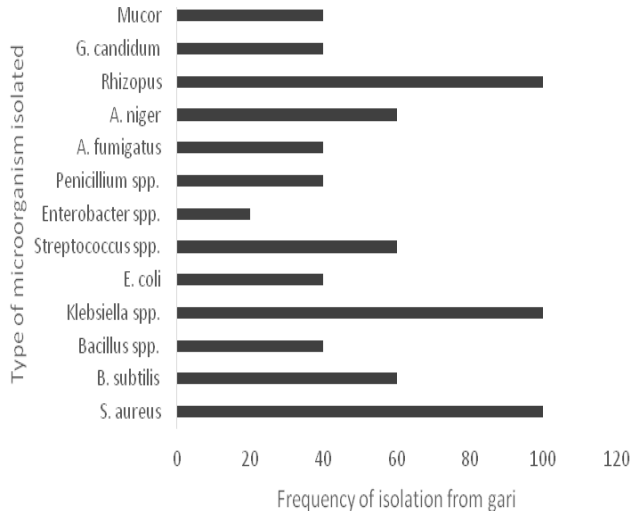

Figure 2: Frequency of isolation of microorganisms from gari samples

Table 3: Biochemical Analysis of bacterial isolates from Gari

	ISOLATES						
	1	2	3	4	5	6	7
Gram Rxn	-R	-R	-R	+C	+SR	+R	+R
Catalase	+	+	+	+	+	+	+
Citrate	+	-	+	+	-	+	+
Motility	-	+	+	-	+	+	+
Starch Hydrolysis	+	+	+	+	+	+	+
MR	-	+	-	-	+	-	-
VP	+	-	+	-	-	+	+
Indole	-	+	-	+	+	-	-
Glucose	A/G	A/G	A/G	A/-	A/-	A/-	A/-
Lactose	A/-	A/G	-/-	A/-	A/-	A/-	A/G
Fructose	A/-	-/-	A/-	A/-	A/-	A/-	A/-
Sucrose	A/G	A/G	A/G	A/-	A/G	A/-	A/-
Xylose	A/-	A/G	A/G	A/-	A/-	A/-	-/-
Probable Organism	*Klebsiella* spp.	*E-Coli*	*Entrobacter* spp.	*Staphy-lococcus*	*Strepto-coccus*.	*Bacillus subtilis*	*Bacillus* spp.

Key: + = positive; - = negative; SR = Short rod; A =Acid production; AG= Acid and Gas production; A/- = Acid no gas

CONCLUSION

Although it is unclear how long the other gari samples had been in the market, except for the Producer (unpackaged) white gari which was newly prepared. Swelling index, water absorption capacity, bulk and packed densities, pH and total titratable acidity were within the general acceptable limits. The vast array of microorganisms (spoilage and pathogenic organisms) isolated in this study may be associated with gari at various points of sale. Some of these microorganisms are responsible for the distinct organoleptic characteristics of the product. On the other hand, the presence of organisms such as *Rhizopus nigricans* can cause the tainting and discolouration of gari while the presence of *S. aureus*, *E. coli*, *Enterobacter* spp. among others cause foodborne diseases or intoxication. Decanting initial gari soak water reduces the microbial load. Therefore, consuming gari as ready to eat without decanting initial soak water especially among vulnerable individuals such as young, old, pregnant and immune-compromised is of public health concern and should be reconsidered.

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